The efficacy of RH-7988 against green peach aphid (*Myzus persicae* (Sulzer)) and cabbage aphid (*Brevicoryne brassicae (L.)*) on cabbages.

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Green peach aphid (*Myzus persicae* (Sulzer)) and cabbage aphid (*Brevicoryne brassicae (L.)*) are pests of brassicas due to their ability to reduce the quantity and quality of yield through direct and indirect damage. Problems with resistance to current aphicides and consumer demand for selective pesticides has led to the need for new aphicides. Towards this end, field and laboratory experiments were set up to compare a novel aphicide (RH-7988) with a currently registered aphicide (pirimicarb), for aphid control in cabbages.

Three field trials were conducted using a 5x5 latin square design. The number, size and species of aphid colonies per plant was recorded at intervals after treatment. In laboratory experiments the LC_{50} and temperature/toxicity relationships of RH-7988 and pirimicarb were determined using aphids treated

on leaf discs. To investigate the residual activity of RH-7988 aphids were caged on to treated cabbages and mortality was assessed 24 and 48 hours after treatment. The toxicity of RH-7988 to several natural enemies was determined by exposing insects to fresh (wet) spray residues.

No phytotoxic effects were recorded on cabbages sprayed with RH-7988 plus surfactants (Triton B1956, Triton AG98, Sunspray 6E and Citowett). RH-7988 was equivalent to pirimicarb in controlling aphids on cabbages in the first 14 days after treatment (aphid populations were <20% that of the control treatment). After this time (up to 35 days after treatment) aphid populations on RH-7988 treated plants were, on average, 38% that of populations on plants treated with pirimicarb.

There was no significant difference between the LC_{50} values of both aphicides when tested against each aphid species. The toxicity of RH-7988 to the two aphid species did not change significantly between 10 and 30^{o}C and were not significantly different to pirimicarb. The aphicides had a significant residual effect (cf. control treatment) on cabbage aphid and green peach aphid for 5 and 10 days after treatment, respectively. RH-7988 also reduced aphid numbers on leaves that emerged subsequent to aphicide application (between 18 and 23 days after treatment). The recommended field rate of RH-7988 (100 g a.i./ha) was more toxic towards the two aphid species (100% mortality) than to several natural enemy species and life stages (corrected means ranging from 0-59.3%).

RH-7988 has good potential to be included as a aphicide for use on brassicas. Excellent control is achieved for periods longer than pirimicarb and RH-7988 is selectively more toxic to aphids than to some of their common natural enemies.

Keywords: aphicide, insecticide, natural enemies, phytotoxicity, pirimicarb, residual activity, selectivity, systemic activity, temperature/toxicity relationship,

TABLE OF CONTENTS

LIST OF TABLES

Table

LIST OF FIGURES

CHAPTER ONE

INTRODUCTION

Aphids are an extremely successful group of insects that are found in temperate and tropical regions of the world, the greater number of species being found in the former.

Over 70 species of aphid have been recorded in New Zealand and most (especially those of agricultural importance) have been accidentally introduced during commercial activities (Valentine, 1967; Miller and Walker, 1984). The aphid species studied in this thesis, green peach aphid (*Myzus persicae* (Sulzer)) and cabbage aphid, (*Brevicoryne brassicae (L.)*) were probably introduced in this manner.

Cabbage was used as the plant substrate in this thesis and is a known host for both species. The two aphids are important pests of cabbages because of their direct (feeding) and indirect (virus transmission) effects on the quality and quantity of crop yield (contamination of the market product). The worst periods for cabbage infestation by these aphids are in spring and autumn when the main aphid flights are occurring. Cabbages and other brassica hosts are grown all year round.

In the past, control of aphids has been readily achieved using systemic insecticides. However, resistance in green peach aphid to these insecticides

has developed and the perception of the consumer towards insecticide use has changed. This has led to reduced insecticide use and the adoption of alternatives to chemicals (cultural methods and biological control). Application of insecticides in this instance, is on the basis of need rather than a regular (calendar) schedule.

Selective insecticides have a narrow range of insect species against which they are active. For this reason they are compatible with biological control methods and programmes to reduce insecticide use, because they enhance natural enemy survival.

The aim of this thesis project was to evaluate the potential of RH-7988 to control aphids on brassicas. Specific objectives were:

(a) to determine the toxicity of RH-7988 to cabbage aphid, green peach aphid and selected natural enemies in laboratory studies;

(b) to define how long RH-7988 was active against aphids in potted plant experiments;

(c) to determine the efficacy of RH-7988 for aphid control in the field and to assess phytotoxic effects on cabbages;

(d) to compare the efficacy against a currently registered selective aphicide, pirimicarb.

CHAPTER TWO

LITERATURE REVIEW

2.1 Aphids as pests

An aphid as an individual has little effect on its host plant. However, with their ability to increase in numbers at a phenomenal rate, aphids can be present in such numbers that they impose a serious nutrient drain on their host plants (Miles, 1989b). Aphids are also vectors of many plant viruses that cause diseases and can lead to serious yield loss.

Aphids may have a considerable effect on yield both directly and indirectly (Wellings *et al*, 1989). Aphid feeding is responsible for most of the direct damage through the removal of nitrogen and carbohydrate and the injection of physiologically active substances in the saliva (e.g., those responsible for gall formation; Miles, 1989b). Indirect damage is mainly attributable to aphid excretions (honeydew which supports black sooty mould) and frass decreasing the photosynthetic efficiency of the host.

2.1.1 Rate of increase in aphids

Aphids are capable of achieving a remarkable rate of population increase (Dixon, 1987). A number of factors contribute to this attribute of aphids: polymorphism, parthenogenesis and the telescoping of generations.

An aphid's life cycle is composed of a number of morphs and a division of labour has developed where each morph has a particular role to perform: dispersal, reproduction or survival (Dixon, 1985a). The resources of each morph are partitioned towards the one major functional role that they play in the life-cycle. With little other requirements on their resources those morphs whose role is reproduction are capable of high reproductive rates.

The majority of aphid species undergo cyclical parthenogenesis; that is; for most of the season the female apterae (wingless) and alatae (winged) will reproduce asexually, producing clones of themselves. Once a year, during autumn a generation capable of sexual reproduction may be produced and eggs will be laid. These eggs overwinter in an undeveloped state and will hatch in spring after completing development.

By being parthenogenetic for most of the year an aphid population is able to increase much more rapidly than it would if using sexual reproduction. By retaining sexual reproduction the genes within the gene pool are mixed and a wide range of genotypes is maintained in the environment.

Rather than waiting until parturition, the embryos of a parthenogenetically reproducing aphid have embryos developing within them and as a result the period between adult moult and the onset of reproduction is shortened. This phenomenon is referred to as the telescoping of generations.

2.1.2 Dispersal of aphids

Dispersal is a further reason for the 'success' of aphids in gaining pest status. The alatae are the life-stage specialised for the role of dispersal and are capable of dispersing great distances on air currents (Roberts, 1987).

To be capable of flight, resources must be partitioned away from reproductive investment. To minimise the drop in reproductive rate, markedly smaller offspring are produced and this increases the total number of offspring per alatae (Dixon, 1985a).

There is an assumed high mortality suffered during dispersal (Dixon, 1985b). To compensate for this, large numbers of alatae are produced (mainly in response to host quality and crowding) and the successful alatae tend to not just settle on one host plant but will maximise dispersal by larvipositing on a number of host plants.

2.1.3 Transmission of viruses

Aphids are important vectors of many plant viruses, the number of plant viruses transmitted by aphids is much larger than the number transmitted by any other comparable invertebrate taxon (Swenson, 1968). This topic has been reviewed by a number of authors: Broadbent (1957), Swenson (1968).

The incidence of plant viruses is closely related to that of their aphid vectors. Broadbent (1957) noted that the fluctuations in numbers of cabbage aphid and green peach aphid had a considerable influence on the incidence of cauliflower mosaic virus and cabbage black ring spot virus in brassica crops in the United Kingdom. Watson and Healy (1953) reported that variation among fields in total numbers of alate green peach aphid trapped throughout a growing season accounted for 83% of the variation in beet yellows virus incidence among sugar beet fields.

2.2 Key aphid pests of brassicas

Cabbage aphid and green peach aphid are the two main pests of brassica crops in New Zealand. False cabbage aphid (*Lipaphis erysimi* (Kaltenbach)) is also found in New Zealand but is generally of minor importance (Butcher, 1984).

2.2.1 Green peach aphid

van Emden *et al* (1969) considered green peach aphid to be outstanding in distribution (found world-wide) and in host plant range (47 host species recorded in New Zealand; Cottier, 1953). Direct damage is incurred on brassica hosts by green peach aphid populations which are commonly found on the senescing leaves, with small populations occasionally being found on the young tissue of the host. Green peach aphid is also a vector of virus diseases to plants from 30 different plant families (including many major crops).

Kennedy *et al* (1962) compiled a list of 182 viruses that green peach aphid was known to transmit.

Green peach aphid has a holocyclic life-cycle; i.e., it exhibits both asexual and sexual reproduction (in those environments where the winter is harsh enough to require it). The eggs of the sexual phase are laid on the primary host, *Prunus persica* and hatching in spring closely coincides with the phenology of the host.

In spring a population explosion occurs initially, because of a rich food supply, but as summer approaches the population begins to decline due to a decrease in the nutrition status of the host plant and the effect of natural enemies (van Emden *et al*, 1969). Associated with the decline in food quality there is a drop in aphid reproduction. The number of alate aphids produced increases due to poor nutrition and crowding and these migrate from the primary host to the secondary hosts.

Green peach aphid will disperse throughout summer (between secondary hosts) as the suitability of the current host declines. Near the end of the summer or in autumn winged gynoparae and males are produced on the secondary hosts and these migrate back to the primary host where the eggs are produced after mating. The eggs hatch in spring and the cycle begins again.

Green peach aphid maintains a choice of overwintering methods (Blackman, 1974). Because of its wide host range, green peach aphid may survive the winter on secondary hosts, but only if the winter is a relatively "mild" one. In this situation, green peach aphid overwinters as apterous, parthenogenetic females which may continue to reproduce throughout the winter and alatae can disperse to new hosts as old hosts decline in suitability.

The critical low temperature for survival of green peach aphid apterae is below 2^{o}C (Adams, 1962), the level depending on cold hardiness. Heie and Peterson (1961), cited by van Emden *et al.* (1969), suggested that a mean temperature greater than 4^{o}C during the three coldest months was required for survival of green peach aphid apterae. Heinze (1939), cited by van Emden *et al.* (1969), considered that the average monthly maximum in winter had to exceed 10^{o}C for green peach aphid apterae survival.

2.2.2 Cabbage aphid

Cabbage aphid is considered by Schepers (1989) to be the most harmful aphid species to brassica crops all over the world. Twenty-nine viruses are transmitted by cabbage aphid and direct damage is suffered by the crop through the aphid's feeding. Tissue is damaged (leaves turn lumpy with curling edges; Schepers, 1989) and pale spots (chlorosis) appear around feeding punctures (Miles, 1989a). The effect of this is to lower the total photosynthetic output of the host.

In New Zealand, cabbage aphid is only found on plant species within the Cruciferae family. The male has been recorded in New Zealand (Cottier, 1953), but the sexual phase of the life-cycle has not been observed. Hughes (1963) noted that parthenogenetic reproduction could continue through the colder part of the year in Australia and that the sexual cycle had largely been suppressed because of this. This may explain the absence of the sexual cycle in New Zealand.

The life-cycle of cabbage aphid is similar to that for non-sexually reproducing green peach aphid, although the cabbage aphid only has the one family of host plants. Peak flights occur in spring and autumn as the aphids migrate to find young highly suitable hosts. At other times during the year alatae may fly in search of new hosts as the occupied host becomes over-populated.

2.3 Control of aphid pests

The control of aphids that are pests of agricultural and horticultural crops can be obtained by a number of means and these can be conveniently grouped into three categories: chemical, cultural and biological control. Integrated Pest Management (IPM) programmes aim at maintaining control of aphid pests by using methods from all three categories.

2.3.1 Chemical control

Prior to the second World War chemical insecticides used to control aphids consisted mainly of nicotine and some arsenic based products (Schepers, 1989). The organochlorines (OC) were the first synthetic insecticides on the market used to control aphids but their persistence in the environment has lead to their widespread withdrawal. Organophosphates (OP) and carbamates introduced less persistence in the environment and many insecticides in these two groups also possess systemic activity. The (synthetic) pyrethroids overcame the instability of natural pyrethrin in the environment and are non-toxic to mammals (Elliot, 1976).

The 1990 New Zealand Agrichemical and Plant Protection Manual (O'Connor, 1990) lists 13 and 17 insecticides for aphid control in forage brassicas and vegetable brassicas, respectively. Of the total of 20 chemicals registered for use against aphids on brassicas 17 are organophosphates, two are carbamates, and one is a mixture of an organophosphate and a pyrethroid. Of these insecticides, 11 have a noted systemic activity (9 OPs and the two carbamates).

Seedling brassicas are the most vulnerable plant stage because their small size means that lower numbers of aphids are required to inflict injury. Early control is also important to protect the seedlings from aphid-transmitted viruses.

Granular insecticides applied to the soil at planting gives long protection against aphid attack (see Schepers, 1989), however, their effectiveness varies with soil humidity and is lower under dry conditions (Suett, 1977; cited in Schepers, 1989). Systemic insecticides with long residual activity are sprayed during the growth of the crop. Closer to harvest insecticides with low persistence are used to avoid problems with withholding periods.

A problem associated with chemical control is the selection for resistance in the pest to the chemical(s) being used. Resistance in green peach aphid to commercial aphicides was first detected in the United Kingdom in glasshouse populations (Needham and Sawicki, 1970). In the field, resistance was reported in green peach aphid from sugar beet crops in the early 1970s (Needham and Devonshire, 1975). Furk (1986) found no evidence of any increase in the incidence of resistant green peach aphid since the previous survey in 1976 (conducted by Sawicki *et al*, 1978), while Devonshire (1989) considered that low levels of resistance are now widespread (in the UK).

The resistance in green peach aphid is characterised by a broad cross-resistance to most aphicides. This is greatest to pyrethroids, least to carbamates, with organophosphates in between the two extremes. In considering the observed cross-resistance patterns, Sawicki and Rice (1978) proposed that applying mixtures of pyrethroids and organophosphates is unlikely to give any benefit in terms of decreased resistance in green peach aphid. ffrench-Constant *et al* (1987) provided evidence to support this

hypothesis; an organophosphate/pyrethroid mixture selected for a resistant variant.

Reversion from resistance to susceptibility has been observed in green peach aphid (Beranek, 1974; Sawicki *et al*, 1980; Bauernfeind and Chapman, 1985). The loss of resistance appeared to be random (Beranek, 1974). ffrench-Constant *et al* (1988) induced the reselection of resistance in reverted sub-clones of green peach aphid by increasing the concentrations of insecticide.

Similar resistance has been noted in green peach aphid populations in both Canada (McClanahan and Founk, 1983) and Australia (Attia and Hamilton, 1978; Attia *et al*, 1979; Hamilton *et al*, 1981).

In New Zealand, Fellowes and Ferguson (1974) reported a broad range of resistance in green peach aphid populations from potato crops in south Auckland. Six organophosphates, two carbamates and an organochlorine all gave inadequate control of green peach aphid while acephate (OP) and BAY 6437 (carbamate) both gave adequate control.

Baker (1978) collected a resistant green peach aphid strain from a Christchurch glasshouse and found it to be highly resistant to three organophosphates with cross-resistance to another two, neither of which had been commercially used against the population. Pirimicarb (carbamate)

adequately controlled this population. A susceptible strain from Levin was readily controlled with all insecticides tested in this trial.

More recently, green peach aphid collected from south Auckland potatoes by Cameron and Walker (1988) proved to be resistant (greater than ten-fold difference between LC50s) to three organophosphates (demeton-S-methyl, malathion and dimethoate), a carbamate (pirimicarb) and a pyrethroid (deltamethrin). The population was tolerant (less than ten-fold difference between LC50s) to methamidophos (OP) and methomyl (carbamate) and susceptible to acephate (OP) and endosulfan (OC).

Furk and Murray (1988) reported that RH-7988 controlled resistant strains of green peach aphid at rates required for control of susceptible strains. In the same trial, higher rates of pirimicarb were required to control the resistant strains. Dewar *et al* (1988) found similar results in their laboratory trials.

Herron *et al* (1990) found that RH-7988 was more toxic than pirimicarb to both susceptible and resistant strains of green peach aphid. However, there was a seven-fold resistance in the resistant strain to RH-7988 (c.f. 9.2 fold for pirimicarb), Herron *et al* (1990) concluded that RH-7988 was not likely to be a commercially viable alternative to currently available aphicides for the control of resistant strains of green peach aphid.

Cabbage aphid is also subjected to selection pressure through direct and indirect applications of pesticides but as yet resistance has not developed in this species (Furk and Roberts, 1985).

2.3.2 Cultural control

Prior to the discovery of synthetic insecticides, cultural methods of insect control were of more importance (van Emden, 1989). By manipulating ordinary agricultural practices and disrupting the life-cycles of pests, the incidence of these pests in the crop is reduced. These methods may reduce the intensity of damage but are unlikely to eliminate it.

The methods involved aim at reducing the immigrant population and reducing the success of those immigrants that arrive in the crop. For example, O'Donnell and Coaker (1975) reduced cabbage aphid infestations by 80 percent primarily by interfering with their immigration into the crop.

2.3.2.1 Resistant plants

Occasionally individual host plants appear to be unsuitable for pest development and populations are not found on these plants. By selecting and breeding for these plants resistance/tolerance to the pest can be introduced.

Dunn and Kempton (1972) selected plants that were not colonised by cabbage aphid in the field and found antibiosis in these plants in the laboratory (direct or indirect affects in terms of survival, growth, development rate, fecundity, etc.; van Emden, 1989) and in the field there was a non-preference of incoming alatae to these clones (plants were unattractive or unsuitable for colonisation or oviposition by cabbage aphid; Auclair, 1989).

However, Way and Murdie (1965) noted that while a glossy variety of Brussels sprout was resistant to cabbage aphid, it had increased susceptibility to green peach aphid. A mechanism that confers resistance to one pest species does not necessarily confer resistance to another.

In the same way that resistant (pest) biotypes are selected for in chemical control, biotypes that overcome the resistance of a plant can be selected for in this situation. Palmer (1960) reported that 'Aphis Resistant' rape was generally resistant to cabbage aphid, but Lammerink (1968) later identified that there were two biotypes of cabbage aphid in New Zealand, one of which attacked 'Aphis Resistant' rape severely.

2.3.2.2 Strip-cropping and mulching

Strips of different crops (short and tall crops, crops with different pest complexes) can be sown in a field to confuse incoming immigrant pests and to discourage movement of pests within the crop.

Tukahirwa and Coaker (1982) found infestations of cabbage aphid on brassicas were reduced by over 60% when intercropping with a taxonomically unrelated plant species (c.f. a pure stand of brassicas). Kenny (1985) intercropped dill (*Anethum graveolens*) (taller) with cabbages (shorter) and obtained a significant reduction in cabbage aphid alatae on the cabbages. Dempster and Coaker (1974) grew cabbages with a background of clover and found that the number of alate cabbage aphid entering the crop was greatly reduced (cf. with a bare soil background).

Mulching is used for the control of non-persistent, stylet-borne viruses (carried by incoming alatae). Aphids are not attracted to plants growing close to white or reflective surfaces (Gibson and Rice, 1989) and so reflective mulches (commonly aluminium foil) can be placed between plant rows, sprayed on to the crop as a powder or placed over the whole crop to 'hide' the crop from the alatae.

2.3.2.3 Trap cropping

Trapping the pest in boundary rows or on other more attractive plants reduces the number of pests entering the crop itself. Subsequent eradication of these pests (and often the hosts that they are on) reduces the population immigrating into the main crop. Matthews (1984) gave examples of trap cropping for various pests in rice, cotton and cucurbits. Matthews noted that in cucurbits

the few melon flies that attack the main crop could be controlled by natural enemies.

2.3.2.4 Timing of sowing

Most pests show some seasonal predictability (van Emden, 1989), i.e., flight and oviposition at specific times. Infestation of the crop by a pest can be avoided or reduced through the timing of sowing. The crop may not be present at a time of peak pest movement/activity or is at an age where it is more tolerant of pest populations.

2.3.2.5 Management practices

The methods in this group are part of crop management. The rotation of crops aims, among other things, at ensuring that crops with incompatible pest complexes follow each other and the field, in theory, is 'clean'. Cultivation and weed control removes volunteer plants which may be hosting pests. Pests from the volunteers may otherwise move on to the crop that is subsequently established in the field. In perennial horticultural crops pruning and mineral sprays in winter can reduce the pest problem in the following season.

2.3.3 Biological control

A species is naturally regulated by the action of its natural enemies. Humans have manipulated this to their own advantage as biological control. The classical example is the application of biological control to an immigrant species that has attained pest status in an invaded habitat due, it is presumed, to the absence of the natural enemies that regulate its numbers in its native habitat (Carver, 1989). Natural enemies of the immigrant species are introduced to encourage regulation of the immigrant's population.

The literature on the principles of biological control is voluminous and the reader is referred to the following references for information on this subject: Samways (1981), Huffaker (1971), van den Bosch (1971) and van den Bosch *et al* (1982).

The only known aphid parasitoid of possible economic importance in brassicas is *Diaeretiella rapae* (M'Intosh). It is a parasitoid of many different aphids but in the field it is generally confined to cabbage aphid and green peach aphid on brassica hosts (McLaren, 1975). Lowe (1968) recorded a maximum of 39.4% parasitism by *Diaeretiella rapae* on cabbage aphid populations at three sites in New Zealand. Theunissen (1989) stated that parasitism of apterous cabbage aphid in cabbage crops could approach 100%, especially later in the growing season.

However, the literature concludes that *Diaeretiella rapae* generally fails to achieve desired levels of control (McLaren, 1975; Kenny, 1985). This may possibly be due to hyperparasitism. High levels of hyperparasitism (*Charips brassicae* (Ashm.), *Lygocerus niger* (How.) and *Pacheneuron* spp.) have been recorded (Lowe, 1968). Early (1984) noted that *Alloxysta infuscata* (Kieffer) is an important hyperparasite of *Diaeretiella rapae*.

Predators that prey on aphids in New Zealand are: ladybirds (adult and larvae), lacewings (adults and larvae) and hoverflies (larvae only). The predators tend to be general feeders and do not restrict their prey to any one species and may prey on a range of families and orders.

Three ladybird species habitually feed on aphids: the eleven-spotted ladybird (*Coccinella undecipuncatata*, Linnaeus), the orange-spotted ladybird (*C. leonina*, Fabricius) and the two-spotted ladybird (*Adalia bipunctata* (Linnaeus)). Coccinellids have many attributes of an effective enemy (Hodek, 1967) but in their disfavour they are not strictly host specific in their feeding and have an unfavourable rate of increase compared to that of their aphid prey. Synchrony of the coccinellids with their aphid prey populations is not always achieved and parasitism can reduce the effectiveness of the coccinellids. Coccinellids on their own are usually unable to check aphid infestations effectively, although they are an important component of natural control (Hodek, 1967).

The Tasmanian lacewing (*Micromus tasmaniae* (Walker)) is a general predator, feeding mainly on a number of aphid species (Early, 1984). Leathwick (1989) concluded that the role of the Tasmanian lacewing was restricted by its low appetite for prey (and the crop management practised). Leathwick (1989) also noted that the lacewing could attack aphids early in their population growth phase and that large numbers of lacewings could be present in the crop (lucerne). New (1975) considered that the tolerance of lacewings to some insecticides enhanced their potential use in integrated control.

There are two native species of hoverfly (Syrphidae) whose larvae feed on aphids: *Melangyna novaezelandiae* (Macquart) and *Melanostoma fasciatum* (Macquart). The adults are attracted to heavy aphid infestations and can suppress larval numbers until late in the season (Early, 1984). Tamaki *et al* (1967) reported that syrphids were the only predators that effectively suppressed green peach aphid populations on peach trees (autumn).

An entomopathogenic fungus, *Entomophthora aphidis*, Hoffman, was reported by Lowe (1968) as being an important mortality factor of cabbage aphid populations in the autumn. A mean of 33% of the population was diseased by this fungus. McLaren (1968 and 1975) did not report any losses of cabbage aphid due to fungal attack.

A problem that would be encountered with biocontrol in vegetable brassicas is that the aphids are not totally removed from the crop. The aphids

contaminate the market product to some extent with their presence, their frass, the remains from the action of the biocontrol agents. This is unacceptable to the consumer of the fresh market product and lowers the value of the yield (low quality yield).

2.3.4 Integrated Pest Management

Cultural and biological control methods, by themselves, do not allow the production of high quality yields in vegetable brassica crops and may not prevent yield reduction in brassicas. Chemicals will control the aphids only as long as they are susceptible. However, resistance is proving to be a problem especially in green peach aphid where broad cross-resistance is well documented and found world-wide. Thus, as Harrewijn and Minks (1989) suggest, it would be sensible to integrate all available techniques; firstly, to prevent or hinder migration of aphids; secondly, to disturb migrants alighting in the crop and thirdly (ultimately), to reduce aphid numbers in the populations that do establish.

Control programmes that aim to achieve this come under the heading of Integrated Pest Management (IPM). Metcalf and Luckmann (1982) give an excellent introduction to the theories behind this concept.

An integral part of an IPM programme is determining a threshold of pest numbers below which there is no economic damage to the crop. The aim of

the programme is to maintain pest numbers below this threshold, therefore, maintaining a reservoir of hosts/prey for the natural enemies of the pest. If numbers rise above the threshold then control measures (e.g., a pesticide application) are implemented to return numbers to below the threshold.

By using control methods other than chemical means the number of chemical applications can usually be reduced. This reduces the selection pressure towards resistant strains of the pest and the chemicals are applied only when required rather than the calendar (regular) sprays that are commonly used otherwise.

Theunissen (1989) reported on research which led to tolerance levels (thresholds) being defined (and tested) for cabbage aphid on three brassica varieties in the Netherlands. The percentage of infested plants varied depending on the growth stage of the crop and the crop variety.

The limitation of Theunissen's programme was that it considered cabbage aphid only. It is unlikely that cabbage aphid was the only pest present in the crop; other insect pests, plant pathogens and weeds should also be considered. A full IPM programme would encompass all pests and their interactions. An obvious interaction is the fact that cabbage aphid is a vector of virus diseases and this is not considered in the programme described by Theunissen (1989). If this had been considered then one would expect the threshold to be lower

early in the growth of the crop to decrease the chances of an aphid carrying a virus into the crop.

2.4 Pesticide selectivity

For the successful integration of insecticides and biocontrol agents (as is required in an IPM programme), there is a requirement for selective insecticides that have a minimal effect on beneficial insects. This definition is what many refer to as physiological selectivity and is apparent when there are marked differences in acute toxicity between organisms (namely, the target pest and its natural enemies) following equivalent doses of the same toxicant (Brooks, 1976).

Another possible means of achieving selectivity is ecological selectivity, or the selective use of insecticides, so that beneficial insects and other non-target insects are less likely to receive a toxic dose and the likelihood of the target pest receiving a toxic dose is maximised. This is achieved through the selection of an appropriate formulation of an insecticide and the rate, timing and placement of this insecticide on the crop (Graham-Bryce, 1977).

It is generally considered that natural enemies are more susceptible to insecticides than their host/prey (Mullin and Croft, 1985). Theiling and Croft (1988) suggested that this is not necessarily the case since many insect families which include species of natural enemies are more tolerant of insecticides than

their pests. However, the research in this area could be considered to be subjective in that many insecticides tested against natural enemies are likely to be those that show low effects on the natural enemies. In addition, certain stages of some natural enemies are more tolerant to insecticides, e.g., the pupal stage of some parasitoids (Croft, 1990).

RH-7988 is reported as being 'a highly selective aphicide' (Murray *et al*, 1988). Diptera, Coleoptera and Lepidoptera are not controlled at doses that provide aphid control, but more importantly, RH-7988 has been shown to be 'safe' (<30% mortality) against a range of natural enemies (a coccinellid, a predatory mite and two hymenopteran parasitoids) and honey bees (Murray *et al*, 1988).

Pirimicarb is one of the most selective synthetic insecticides registered for field use (Croft, 1990). Pirimicarb had an average toxicity rating of 2.99 (where a rating of 3 = 10-30% effect) to arthropod natural enemies in the SELCTV database (complied by Theiling and Croft, 1988).

The selectivity of pirimicarb is well documented in the literature (Brown et al, 1983 (predators); Franz *et al*, 1980 (natural enemies); Helgesen and Tauber, 1974 (natural enemies); Kennedy and Oatman, 1976 (parasites)) and has been reviewed by Croft (1990). Delorme (1976) concluded that pirimicarb had medium toxicity to the aphid parasitoid, *Diaeretiella rapae*.

2.5 Sampling methods

2.5.1 Design of field experiments

The general rule for field experiments is that they should provide maximum information with minimal effort (Nelson, 1976; Thompson and Wheatley, 1977).

Well designed field experiments have a number of similar characteristics:

(a) The design is generally balanced (the same number of replicates for each treatment). This an advantage in the analysis and interpretation of results (Nelson, 1976).

(b) Treatments are randomly assigned to plots and this assures that a treatment will not continually be favoured (Thompson and Wheatley, 1977). Fisher (1951) stated that the random choice of treatments is a complete guarantee of the validity of the test of significance (of the results).

(c) The experiment will have enough replicates to provide sensitivity and precision to the results but will not have so many so as to make the experiment "too large" in terms of management and sampling.

Randomised block designs are the most common field experiment design (Nelson, 1976; Thompson and Wheatley, 1977), while split-plots, latin square

and completely randomised design are other experimental designs that are frequently used.

The advantage of the latin square design is that it provides error control in two directions (Cochran and Cox, 1957; Nelson, 1976). A restriction on this design is that the number of treatments does not usually exceed twelve since the number of replications must equal the number of treatments (Cochran and Cox, 1957). Also randomisation in this design is more difficult because each column and row contains a complete replicate.

Thompson and Wheatley (1977) note that random placement of untreated plots within each block of an experiment will only be satisfactory when variation in infestation (and other relevant factors) are random too. If infestation of the subject pest was known not to be random, Thompson and Wheatley suggest using semi-systematic designs in which the untreated plots are systematically deployed and the (remaining) treatments allocated at random among the remaining plots.

Aphid flight is random (Blackman, 1974; Dixon, 1985b) and the infestation of alatae into a brassica crop is random. This precludes Thompson and Wheatley's requirement for a semi-systematic design and allows the use of the latin square design.

2.5.2 Sampling aphids in field brassicas

The distribution of green peach aphid and cabbage aphid in brassicas is well documented by: Dunn and Kempton (1971) and van Emden and Bashford (1969), on Brussels sprout; Trumble (1982), on broccoli. Green peach aphid prefers the oldest leaves, while cabbage aphid prefers the youngest leaves.

Trumble (1982) considered that whole plant samples were tedious to assess and suggested that aphid density could be reliably and effectively estimated by using leaf counts. Whole young plants would be sampled but with more mature plants only older leaves would be sampled where green peach aphid was the dominant species present and young leaves where green peach aphid and cabbage aphid co-existed.

This method may be useful in monitoring aphid presence in a crop for an IPM programme. A more accurate estimate of aphid numbers and the effect of aphicides on the aphid populations was required so whole plant (above ground) samples were used in this project.

2.5.3 Insecticide bioassays

Insects differ in their susceptibility to a given insecticide and bioassays are used to determine the appropriate dosage for individual pest species (Matthews, 1984). The response of individuals of a species to an insecticide is

variable. Finney (1971) showed that this variability is greatest at the extremes of response. For this reason the value that is commonly quoted for a species is the (lethal) dose or (lethal) concentration that causes a response (mortality) in 50% of the population (LD_{50} and LC_{50}, respectively).

A number of factors can affect the outcome of a bioassay. These have been discussed by Matthews (1984) and Potter and Way (1958) and are listed here under three headings:

(a) intrinsic factors - species specificity, (life) stage specificity and age, sex and size.

(b) extrinsic factors - temperature, humidity, food, population density and illumination.

(c) experimental factors - standardised methods, handling of insects and defining mortality.

Because of the variability in the reaction of test subjects, another feature of bioassays, noted by Finney (1971), was the consequent impossibility of reproducing, at will, the same result in successive trials however carefully the experimental conditions are controlled. For this reason, it is extremely important that the above factors are controlled so that the experiment can easily be repeated, although the same results may not be exactly reproduced.

The definition of mortality needs to be clearly stated both in the experiment plans and any publications concerning the experiment. Measurements based

on the mortality of the organism render the most unambiguous results since the aim of an insecticide is to kill the insect, death is irreversible and is equivalent in all insecticide groups and in all insect life stages (Bánki, 1978). Bánki (1978) also noted that it is not always easy to establish that death has occurred in the case of small insects and suggested the measurement inhibition instead (which is reversible). For example, measuring the lack of coordinated movement rather than a total lack of movement may be more appropriate.

A bulletin published by the Food and Agriculture Organisation (FAO) on bioassays (Anon, 1979) states that the criterion of death varies and once chosen for an experiment must be maintained. The suggested method in this publication is to gently prod the insect; if it falls over and is unable to get up, it can be presumed dead. This method would not work with insects that "play dead" when disturbed. Obviously the method may need to change as the circumstances change. In the literature the definition of mortality is varied. Aphids were considered dead when there was no voluntary movement after prodding by Kumar and Chapman (1984), Furk and Roberts (1985), McLeod (1987) and Furk and Murray (1988); Syrett and Penman (1980) relied upon some form of normal locomotory behaviour (or lack of it) in lucerne aphid and two predators in their work to define mortality while McClanahan and Founk (1983) used the FAO definition, above.

Obviously, mortality, as defined in bioassays, is not always true mortality although in most cases the insects end up dying, given time. One could argue

rather semantically that the term mortality (and thus, LC_{50} and LD_{50}) should not be used in these cases, but given the wide range of different methods and the difficulty in reproducing results, the technically incorrect use of the term is not of great importance.

Syrett and Penman (1980) recorded 48 hour data for bioassays on lacewings and ladybirds but used the 24 hour data for the lucerne aphid. They justified the choice of the shorter period for the aphids because of their shorter lifespan. Kay (1979) recorded and presented both 24 and 48 hour data. The LD_{50} fell over this time period for all of the nine insecticides tested as did the difference between fiducial limits.

Galley (1968), in a study on bioassay techniques, found that a modified leaf disc technique increased in sensitivity asymptotically with time and was nearly three times as great after 48 hours compared to 24 hours. Galley also noted that variability between replicate treatments increased either side of the 48-72 hour time period and suggested that this interval represents an optimum between uptake of the toxic compound and its metabolic degradation in significant quantities within the discs.

The general opinion of the literature is that 48 hour data gives more reliable results. However, there are cases where the use of other time periods can be justified and it is up to the researcher to make the choice and clearly define that choice.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Products tested

RH-7988 (ethyl(3-tert-butyl-1-dimethyl carbamoyl-1H-1,2,4-triazol-5-ylthio) acetate) has a solubility (technical grade) of less than 1% in water, and is soluble in methylene chloride and ethyl acetate. The acute oral and dermal LD_{50} values of RH-7988 for rats is 50-200 mg/kg and 5000 mg/kg, respectively. RH-7988 was available as an experimental 25% wettable powder formulation and a 480 g/l emulsifiable concentrate formulation. In the first two field experiments the latter was used. At the request of Rohm and Haas New Zealand Ltd., the formulation used for the remaining experiments was changed to the wettable powder.

Pirimicarb (2-dimethylamino-5,6-dimethyl-4-pyrimidinyl dimethyl carbamate), trade name Pirimor (ICI), has a solubility of 2.7 g/l in water (aqueous solutions tend to be unstable) and is soluble in most organic solvents. The acute oral and dermal LD_{50} values of pirimicarb for rats is 147 mg/kg and 600 mg/kg, respectively. Pirimicarb is marketed as a water dispersible granule (500 g/kg) and it was this formulation that was used in these experiments.

Triton B1956 surfactant was used in all experiments to improve spray retention. Triton B1956 contains 77% modified phthalic glycerol alkyd resin

and 23% 1,1,1 trichloroethane and has an acute oral LD_{50} of 2200 mg/kg against rats.

Citowett (1000 g/litre alkylaryl polyglycol ether), Triton AG98 and Sunspray 6E (970 ml/litre mineral oil, 92% minimum unsulphonated residue content) surfactants were included in Field Experiment I to test for phytotoxic effects on cabbages.

3.2 Field Experiments

3.2.1 Experiment design

Three field experiments were carried out at Lincoln University over an eighteen month period. Cabbage seedlings (*Brassica oleracea* cv. 'Golden Acre' Merit Strain) were transplanted into plots in a latin square design. This allowed for a reduction in the errors through the removal of the within row (NW-SE orientation) and column (SW-NE orientation) differences. Each plot contained 49 plants at 0.5m intervals (both within and between rows) and was separated from adjacent plots by a 1m buffer. Plants in the outside rows/columns of each plot were excluded from the sampling plan and those that were sampled were randomly selected from the inner 25 plants of each plot. Each experiment had five treatments, replicated five times.

Management practices such as: irrigation, weed control and applications of *Bacillus thuringiensis* (Thuricide HP WP, 0.5-1.0 kg/ha) for control of lepidopteran pests were carried out as required, in each of the field experiments.

3.2.2 Sampling methods

Complete counts of aphids on the plants would have been too time consuming and was not really warranted in relation to the experimental objectives. Thus, a sampling method that gave an estimate of the numbers of aphids present per plant was required. In this study, whole plant samples were considered more appropriate than leaf samples, given the spatial separation of the two aphid species on the cabbage host. The number of colonies per plant was counted and recorded, while colony size was categorised into a range of size classes.

3.2.3 Field Experiment I

This experiment was set up to establish whether RH-7988 alone or in combination with a range of surfactants and rates of surfactant, was phytotoxic to cabbages.

Cabbage seedlings were transplanted in late November, 1987, including three guard rows planted at the top and bottom of the experiment. Four commercial surfactants were applied with RH-7988, late January, at varying rates (Table

4.1), to split-plots. Treatments were applied through a precision plot sprayer with a 1.5m boom at 345 kPa and a spray rate of 500 l/ha. At the time of treatment the cabbages had heads that were a quarter to a half formed.

Phytotoxic damage to the plants was assessed 11 days after treatment (DAT) and scored on a scale of 0-5 (where 0 = no damage). At the same time (to assist with the design of subsequent field experiments) the efficacy of RH-7988 was initially investigated by assessing the presence/absence of aphids on eight plants from each plot (aphids numbers were not sufficient to do a full sample as described for the following two experiments).

3.2.4 Field Experiment II

Following the analysis of the presence/absence data gathered in the first experiment, a second experiment was designed which aimed to investigate the efficacy of several rates of RH-7988 compared with a currently registered aphicide for control of aphids on cabbages.

Seedling cabbages for this experiment were transplanted in mid March, 1988. Five treatments (RH-7988 at 50, 100 and 200 g a.i./ha, pirimicarb at 250 g a.i./ha and a control) were applied 25 days after transplanting, using a precision plot sprayer with a 3m boom operated at 345 kPa to deliver a spray rate of 500 l/ha. All treatments included the surfactant Triton B1956 at 0.03% and the cabbage plants had, on average, three true leaves.

Five plants per plot were randomly selected and samples were taken 1 and 10 days prior to treatment and 2, 7, 14, 22 and 35 DAT. At each sample date the plants were divided into three strata:

1 - oldest leaves beginning to senesce and losing their colour, leaf orientation below horizontal;

2 - mature, fully expanded leaves, orientation horizontal or above horizontal;

3 - young, expanding leaves, including the outer two layers of the heart, if present.

This was done in acknowledgement of the preference exhibited by green peach aphid for the oldest cabbage leaves, and by cabbage aphid for the youngest leaves (Trumble, 1982).

The following was recorded for each plant: number of leaves per stratum, number of aphid colonies per leaf, number of alate aphids per leaf and the height of the plants (mm). For each leaf the mean size (1, <10, <50, <100, <200, >200) of the colonies of aphids on that leaf was noted. Natural enemies of aphids that were observed in the crop at sampling were also noted.

Five plants per plot were randomly selected for harvesting in mid- August and total fresh weights and marketable fresh weights (old outer wrapper leaves removed to produce the consumer product) were recorded.

3.2.5 Field Experiment III

From the analysis of results from the second experiment a narrower range of rates of RH-7988 was selected for further efficacy studies compared with pirimicarb.

Seedlings for this experiment were transplanted mid-September, 1988. The five treatments of the main experiment were: RH-7988 at 25, 50, 75 and 100 g a.i./ha. The yield data from the second field experiment indicated that RH-7988 could have an effect on the yield of cabbages, thus higher rates were applied (RH-7988 at 150, 200 and 300 g a.i./ha; all including 0.03% Triton B1956) to small observation plots to investigate this effect. Two further treatments were applied (RH-7988 at 100 g a.i./ha without surfactant, and one untreated plot) to small observation plots. The observation plots had two replicates.

All treatments were applied 30 days after transplanting, through a precision plot sprayer, using a 3m boom operated at 345 kPa to deliver a spray rate of 500 l/ha. All treatments of the main experiment included 0.03% Triton B1956 surfactant. At the time of treatment the cabbage plants had four fully expanded leaves, slightly more than at the previous experiment because inclement weather had delayed spraying.

With the exception of three changes, measurements that were described for the previous experiment were taken at each sample date: 2, 7, 14, 20 and 26 DAT (including the fourth and fifth treatments of the minor experiment; three plants per replicate). The changes made to the method of sampling in this experiment were:

(a) the species forming the aphid colonies were differentiated;

(b) the size of every aphid colony was recorded (rather than an average size per leaf);

(c) the plants were not divided up into the three strata defined in the previous experiment (data collected in previous experiment was not sufficient to warrant the time input).

Five cabbages per plot in the main experiment and three cabbages per plot from the observation plots, were harvested in late December and total and marketable fresh weights were recorded as per Field Experiment Two. The dry weights of marketable heads of three cabbages per plot (main and minor experiment) were also recorded.

3.2.6 Analysis of field experiment data

For each field experiment an analysis of variance was carried out (using SAS statistical package; Ray, 1982) on all data sets except the size of aphid colonies which were analysed using a Chi-square analysis. Least significant difference

(LSD) values were also calculated and if the main effects were significant (F ratio $p<0.05$) then comparisons were made between the RH-7988 treatments and the control and then the pirimicarb treatments. Comparisons (using LSD values) within the RH-7988 treatments and between the control and pirimicarb treatments were not made.

3.3 Laboratory Experiments

3.3.1 Insect rearing

3.3.1.1 Aphids

Cabbage aphid and green peach aphid colonies were reared in insectaries on cabbage plants. Apart from a light source to provide 16 hours light per day, the environment was not controlled. Seedlings were reared separately and were introduced to the colonies as required. The colonies originated from unsprayed field brassicas. As aphids were required for experiments cabbage leaves would be removed and taken to the laboratory. Aphids of a similar size were used in all laboratory experiments, namely apterous adults or penultimate instar nymphs.

3.3.1.2 Lacewings

Adult lacewings, *Micromus tasmaniae* (Walker), were caught with an aspirator in the previously mentioned brassica crops. The adults were caged in perspex boxes (220 x 110 x 80mm, 2 x 25mm diameter vents) for two to three days to ensure that all females had been mated. Moist filter paper and abundant aphids were provided.

Groups of four adults were transferred to petri dishes, again, containing moist filter paper and an abundant supply of aphids, along with a piece of fine terylene gauze as an oviposition substrate (although eggs were often laid on the top, sides and bottom of the dishes, on the filter paper and on aphid exuviae). The pea aphids (*Acyrthosiphon pisum* (Harris)) used for feeding the lacewings were reared on broad beans (*Vicia faba*) in an insectary.

Lacewing larvae were reared in petri dishes and kept at 15^{O}C. Pea aphids were supplied in abundance and scraps of netting provided refuges for the larvae. Filter paper in the dishes was kept damp. Third instars were used for insecticide bioassays and were removed from the dishes when required.

3.3.1.3 Ladybirds

Adult ladybirds caught in the field were predominantly the elevenspotted ladybird, *Coccinella undecimpunctata* Linneaus. Efforts were made to start a

colony in the laboratory using methods similar to those described for the lacewing colony. Failure to induce the females to oviposit meant that a colony was not established. Insecticide bioassays were therefore conducted with adults that were collected from the field (J. Early, Prices Valley) and in this instance the predominant species used was the orangespotted ladybird, *Coccinella leonina* Fabricus.

3.3.1.4 Hymenopteran parasitoids

Diaeretiella rapae (M'Intosh), a parasitoid of the cabbage aphid, became established in one of the cabbage aphid colonies. Adult parasitoids from this colony were used for insecticide bioassays. Initially some hyperparasites of *D. rapae* (*Alloxysta infuscata* (Kieffer)) were inadvertently included in early experiments. Once identified this species was removed from the colony to promote development of *D. rapae*.

3.3.2 LC_{50} determinations

The insecticide bioassays were set up with as described in a FAO plant protection bulletin (Anon, 1979) with the following alterations:

(a) 2 ml of insecticide solution was applied in a Potter tower at 103 kPa (1 ml at 200 kPa recommended);

(b) the settling time was 15 seconds (5 minutes recommended);

(c) both the leaf discs, the agar substrate and the aphids were sprayed with the insecticide solution (the recommendation was for only the leaf discs to be sprayed).

These changes were made to make the conditions of the laboratory experiments more comparable to the field situation.

The bioassays were conducted in petri dishes. Four (17mm diameter) cabbage leaf discs were laid on agar (5% w/v) and five aphids placed on each disc. The dish and its contents was then sprayed with the predetermined insecticide concentration.

After spraying, five aphids were confined on each disc by means of a rubber ring (19mm diameter, 10mm depth) with one end sealed with fine terylene gauze. The aphids were maintained at 20^{o}C and 16:8 hours light:dark. Counts of the number of aphids that were moribund (responded to prodding, but were not displaying normal activity) and dead (no response to prodding) were made 24 and 48 hours after treatment.

Dose-response studies were carried out with both RH-7988 and pirimicarb. Initially, the same broad range of concentrations was used for each aphid species. In successive experiments the range of concentrations was narrowed to more accurately estimate the LC_{50}. Data from the dose-response experiments were analysed using POLO (Russell *et al*, 1977). The experiment

was concluded when the calculated G value (a statistic used in the calculation of fiducial limits; Finney, 1964), for the 24 hour data, was less than 0.5. If the G value is greater than 0.5 the POLO programme will not calculate the fiducial limits.

3.3.3 Effect of post-treatment temperature on toxicity

Using the LC_{50} values determined from experiments outlined in the previous section, the effect of temperature on toxicity was studied. The post-treatment temperatures selected were: 10, 15, 20, 25 and 30^{o}C. The experimental procedures used were the same as those described for the dose-response studies except that the dishes were maintained at one of the above temperatures (16:8 light:dark).

This experiment was replicated over time because of the handling-time and the numbers of aphids required. Three replicates of the treatments at each temperature were carried out initially, with sets of two replicates of any one temperature run subsequently to reduce variability (expressed by the 95% confidence interval). Results were pooled for analysis.

Data from the post-treatment temperature experiments was analysed using Minitab (Ryan *et al*, 1982). A regression was fitted to each data set and 95% confidence intervals were calculated for each datum point.

3.3.4 Residual activity against aphids

The aim of these experiments was to evaluate the extent to which RH-7988 exerts control on cabbage aphid and green peach aphid after spray application.

An experiment was carried out in an insectary for each of the two aphid species. Seedlings were reared in separate pots and had an average of three true leaves when sprayed. Using a precision plot sprayer, the treatments applied to those plants were: RH-7988 at 25, 50, 75, 100 and 200g a.i./ha, pirimicarb at 250g a.i./ha and a water-sprayed control. All treatments included Triton B1956 at 0.03% and each treatment was replicated five times.

Five aphids were caged on to a leaf (the same leaf at each date, where possible) of each plant 1, 5, 10, 20 and 30 DAT. All leaves had received direct spray treatment. Counts of dead and moribund aphids were made 24 and 48 hours after being caged on the leaves. To investigate systemic activity of the aphicides, aphids were also caged and counts made on leaves that emerged subsequent to spraying 18 and 27 DAT for cabbage aphid and 23 and 27 DAT for green peach aphid.

Data from this experiment were analysed using the SAS statistical package (Ray, 1982). $LSD_{0.05}$ values were calculated to enable comparisons between the RH-7988 treatments and the pirimicarb treatment.

3.3.5 Toxicity to natural enemies

By studying the toxicity of RH-7988 to natural enemies of aphids an appreciation of the selectivity of the aphicide could be gained.

RH-7988 at one- and two-times the recommended field rate (0.2 mg a.i./ha) was applied to a selection of natural enemy species and life stages. Petri dishes (bottoms and lids) were treated under a Potter tower (2ml applied at 103 kPa) and natural enemies were transferred into the dishes immediately afterwards. Adult lacewings and Hymenoptera were anaesthetised with CO_2 for ease of handling. The lacewing larvae were given fresh aphids each day to avoid cannibalism.

Counts of mortality and those that were moribund were made 24 and 48 hours after treatment, during which time the natural enemies were maintained at $20^{o}C$.

The standard error of the mean was calculated for each data set using Minitab (Ryan *et al*, 1982).

CHAPTER FOUR

RESULTS

4.1 Field Experiments

4.1.1 Field Experiment I

This experiment tested RH-7988 and a range of surfactants and rates of surfactants for phytotoxic effects on cabbages.

No combination of RH-7988 and surfactant produced severe phytotoxic effects on the cabbages (Table 4.1). However, there was some browning (necrosis) of the outer leaves of the cabbage heads and this increased the phytotoxicity scores. This injury was uniform across the experiment and was possibly due to intense sun and/or, moisture stress.

The higher rate of Sunspray 6E produced the highest phytotoxicity score (2.25) which was significantly higher ($p < 0.05$) than the scores for all other treatments. The importance of this result was diminished by the fact that the control treatment produced the second highest score (2.10) and this was also significantly greater than some of the lower-scoring treatments. The higher rate of Triton AG98 was the lowest scoring treatment (1.85).

Table 4.1: Phytotoxicity injury assessment (0-5, where 0=no effect) and aphid presence on cabbages (11 days after treatment)

Treatment	Phytotoxicity score	Percentage of plants with aphids
RH-7988* + no surfactant	1.95	0
RH-7988 + 0.025% Citowett	1.93	12.5
RH-7988 + 0.03% Sunspray 6E	1.90	0
RH-7988 + 0.06% Sunspray 6E	2.25	2.5
RH-7988 + 0.03% Triton B1956	1.88	0
RH-7988 + 0.06% Triton B1956	1.90	0
RH-7988 + 0.03% Triton AG98	1.95	0
RH-7988 + 0.06% Triton AG98	1.85	2.5
Control	2.10	72.5
SEM	0.177	3.76

* - applied at 100g a.i./ha in all treatments

Cabbage aphid was the predominant aphid species that was present in this experiment. All treatments had a significantly lower proportion of aphid-infested plants (Table 4.1). Excluding the control, all other treatments containing RH-7988 were not significantly different from each other except for the Citowett treatment. The plants with aphids present in the Citowett treatment were all from one replicate and probably had a higher presence due to a spraying error.

4.1.2 Field Experiment II

The efficacy of RH-7988 was compared to a currently registered aphicide in this experiment.

One day prior to spray application there was no significant difference between treatments ($p > 0.05$) in the number of aphid colonies per plant (Table 4.2). By contrast there were significant differences ($p < 0.05$) between treatments on all sampling dates after treatment.

The number of aphid colonies per plant in the pirimicarb treatment was not significantly different to the three RH-7988 treatments 2 days after treatment (DAT). All three RH-7988 treatments had significantly fewer aphid colonies per plant compared to pirimicarb in samples taken 23 and 35 DAT, as did RH-7988 at 100g a.i./ha 7 DAT and RH-7988 at 200g a.i./ha 7 and 14 DAT.

Table 4.2: The number of aphid colonies per plant in the aphicide treatments as a percentage of the control treatments (Field Experiment II)

Treatment (g a.i./ha)		Days after treatment -1	2	7	14	23	35
RH-7988	50	92.6	8.5	9.6	14.0	27.5	37.3
	100	87.0	3.7	3.6	13.9	27.9	42.2
	200	113.0	2.4	2.8	0.0	10.4	31.0
pirimicarb	250	103.3	7.3	17.9	17.2	42.2	60.4
$LSD_{0.05}$		NS	15.88	11.69	12.05	10.10	12.35
SEM		8.58	5.64	4.14	4.28	3.59	4.38
n		75	125	125	125	125	125

The trend in the number of colonies per plant is shown in Fig 4.1. After spray treatment the number of colonies in the control treatment dropped. This effect did not last, however, and the number of colonies in the control treatment rapidly increased to approximately 10 colonies per plant and remained at this level for the rest of the experiment.

From 7 DAT onwards the pirimicarb treatment had more colonies per plant than all three RH-7988 treatments. RH-7988 at 200g a.i./ha consistently had fewer colonies per plant than all other treatments, while the other two RH-7988 treatments had similar numbers of colonies throughout the experiment.

Of the six sampling dates pre- and post-treatment, only 2 DAT and 7 DAT had significant treatment differences ($p < 0.05$) in the number of alatae per plant (Table 4.3). All aphicide treatments had significantly fewer alatae per plant than the control on both of these sampling dates. RH-7988 200g a.i./ha 7 DAT had significantly fewer alatae per plant than pirimicarb; all other RH-7988 treatments were not significantly different to pirimicarb on either sampling dates.

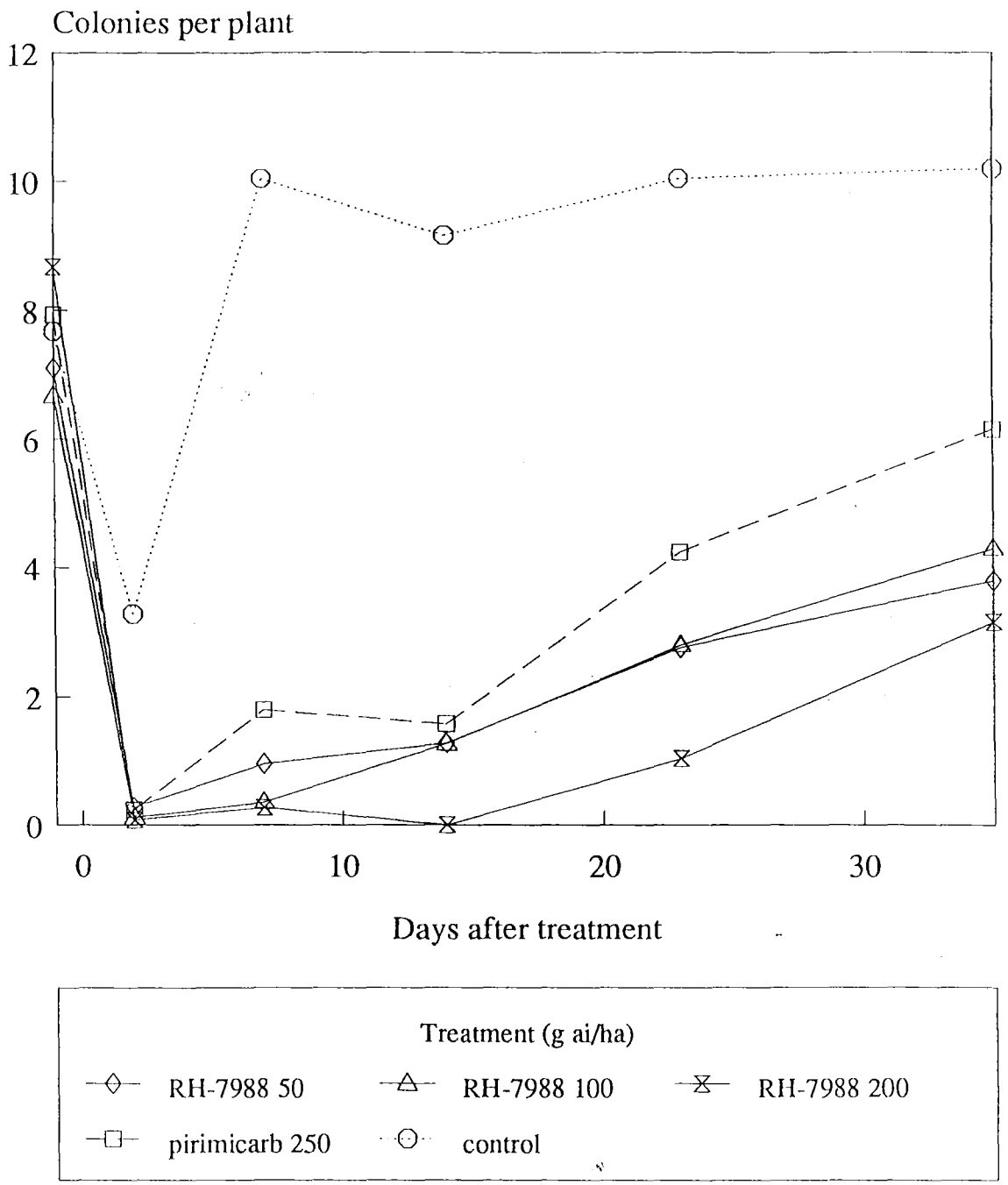

Figure 4.1: The mean number of aphid colonies per plant on cabbages in Field Experiment II.

Table 4.3: The number of alatae per plant in the aphicide treatments as a percentage of the control treatment (Field Experiment II)

Treatment (g a.i./ha)		Days after treatment -1	2	7	14	23	35
RH-7988	50	121.5	39.4	63.3	100.0	120.0	156
	100	80.3	34.8	57.8	103.0	105.0	150
	200	139.1	31.8	37.5	86.6	125.0	148
pirimicarb	250	107.9	48.5	68.0	150.6	107.5	82
$LSD_{0.05}$		NS	35.00	25.29	NS	NS	NS
SEM		25.56	12.42	9.00	16.04	20.56	24
n		75	125	125	125	125	125

The treatments had a significant effect ($p<0.05$) on the total fresh weight of the cabbages (Table 4.4). The total fresh weight of cabbages in the two higher RH-7988 treatments (100g and 200g a.i./ha) were significantly lower than both pirimicarb and the control treatments.

Significant treatment effects were recorded in the market fresh weights of the cabbages ($p=0.0004$). RH-7988 applied at 200g a.i./ha caused a significantly lower market yield than either the control or pirimicarb treatments (Table 4.4). The yield of cabbages sprayed with RH-7988 at 100g a.i./ha was not significantly different to the control but was lower than the market yield produced under the pirimicarb treatment. The market yield of cabbages treated with RH-7988 at 50g a.i./ha were significantly greater than the control treatment but significantly less than pirimicarb treatment.

The data on the size of aphid colonies collected in this experiment were not sufficient for analysis by Chi-square (too many zero counts were recorded).

Table 4.4: Total and market fresh weights of cabbages in the aphicide treatments as a percentage of the control treatment (Field Experiment II)

Treatment (g a.i./ha)		Total fresh weight	Market fresh weight
RH-7988	50	117.4	119.0
	100	99.4	93.2
	200	86.8	73.2
pirimicarb	250	120.0	127.1
$LSD_{0.05}$		19.82	25.54
SEM		7.08	9.12
n		125	125

4.1.3 Field experiment III

A further efficacy study was carried out using a narrower range of RH-7988 concentrations.

There were no significant differences between treatments immediately prior to application of treatments. A mean of 8.13 aphid colonies per plant was found in a sample of one replicate at this time.

Although there was a significant treatment effect ($p < 0.05$) on the number of green peach aphid colonies per plant at every sample date after treatment (Table 4.5a), the number of green peach aphid colonies on plants sprayed with any of the four RH-7988 treatments were not significantly different to those on the pirimicarb treated plants at 2 and 26 DAT. RH-7988 at 100 g a.i./ha had significantly fewer green peach aphid colonies per plant than the pirimicarb treatment 7, 14 and 20 DAT, as did RH-7988 at 75 g a.i./ha 14 DAT.

Table 4.5: The number of green peach aphid and cabbage aphid colonies per plant on cabbages (Field Experiment III)

(a) green peach aphid

Treatment (g a.i./ha)		Days after treatment 2	7	14	20	26
RH-7988	25	3.00	12.36	15.00	13.55	5.22
	50	2.57	9.91	12.76	9.24	6.02
	75	1.36	8.40	9.32	11.98	4.82
	100	0.40	4.56	7.96	2.21	3.62
pirimicarb	250	1.48	10.56	14.20	11.87	3.62
LSD		1.563	3.154	2.806	3.738	2.582
SEM		0.558	1.127	1.002	1.335	0.925
n		125	125	125	50	50

(b) cabbage aphid

Treatment (g a.i./ha)		Days after treatment 2	7	14	20	26
RH-7988	25	0.64	1.16	4.12	3.25	1.2
	50	0.64	1.77	3.04	0.35	0.1
	75	0.32	0.70	2.60	2.15	1.3
	100	0.32	0.44	2.32	0.85	0.2
pirimicarb	250	0.88	2.08	4.72	4.05	2.3
LSD		NS	0.903	1.508	2.681	1.21
SEM		0.228	0.323	0.539	0.957	0.43
n		125	125	125	50	50

Apart from the sample 2 days after treatment, there was a significant treatment effect ($p<0.05$) on the number of cabbage aphid colonies per plant at every sampling date after treatment (Table 4.5b). RH-7988 at 100g a.i./ha had significantly fewer cabbage aphid colonies per plant than pirimicarb 7, 14, 20 and 26 DAT. This was the case for RH-7988 at 75g and 50g a.i./ha on sample dates 7 and 14 DAT and 14, 20 and 26 DAT, respectively.The trends in the number of colonies per plant were similar for both treatments and species (Figs 4.2a and b).

The number of colonies per plant increased bewteen 2 DAT and 14 DAT and decreased bewteen 14 DAT and 26 DAT.

For green peach aphid (Fig 4.2a) the pirimicarb treatment and the two lower rates of RH-7988 (25 and 50g a.i./ha) had a similar number of colonies per plant. Apart from the samples 2 and 26 DAT RH-7988 at 100g a.i./ha had significantly fewer aphid colonies per plant than pirimicarb.

Pirimicarb had more cabbage aphid colonies per plant at every date after treatment (Fig 4.2b) and this was significantly greater than RH-7988 at all dates except 2 DAT.

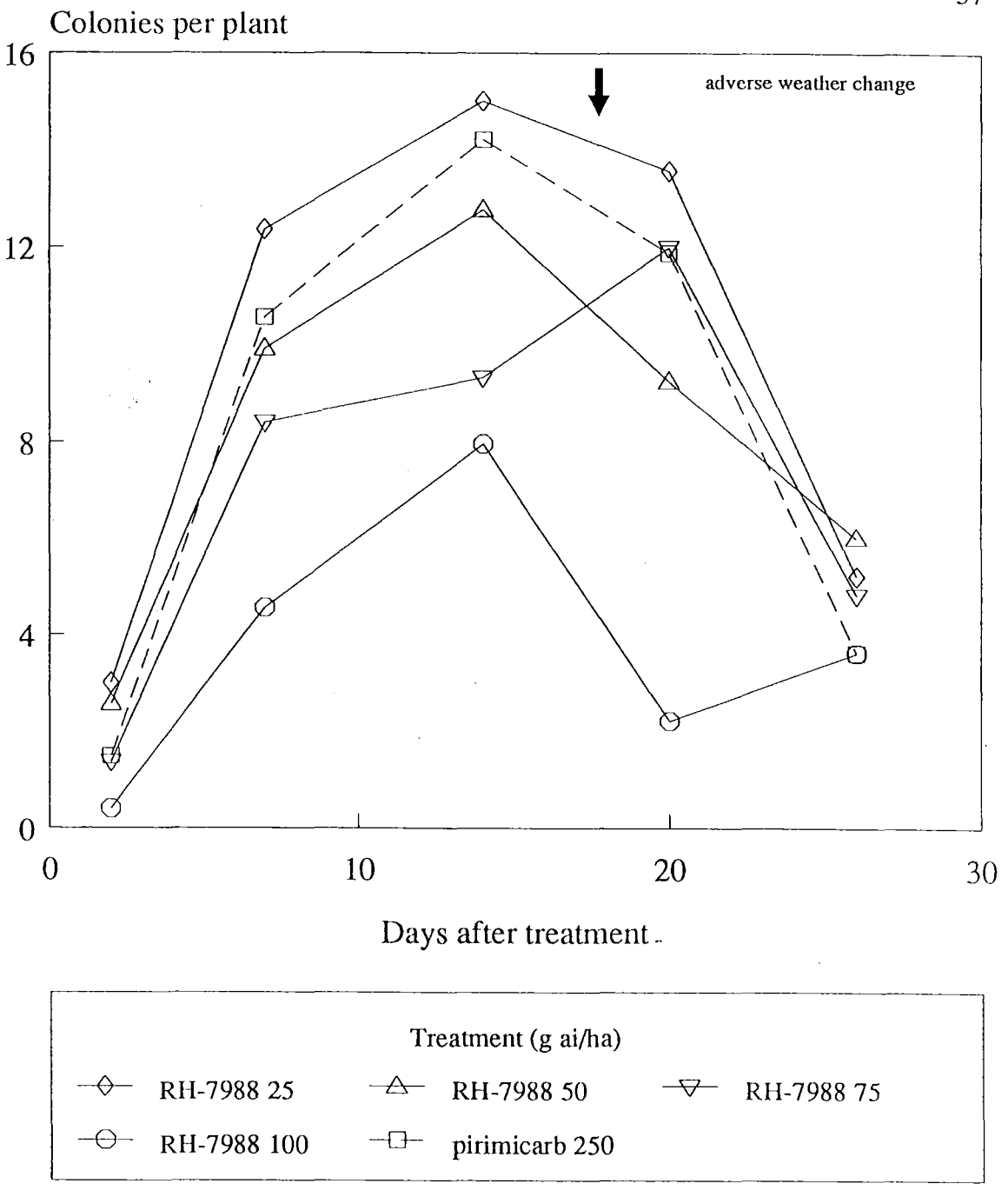

Figure 4.2a: The mean number of green peach aphid colonies per plant on cabbages in Field Experiment III.

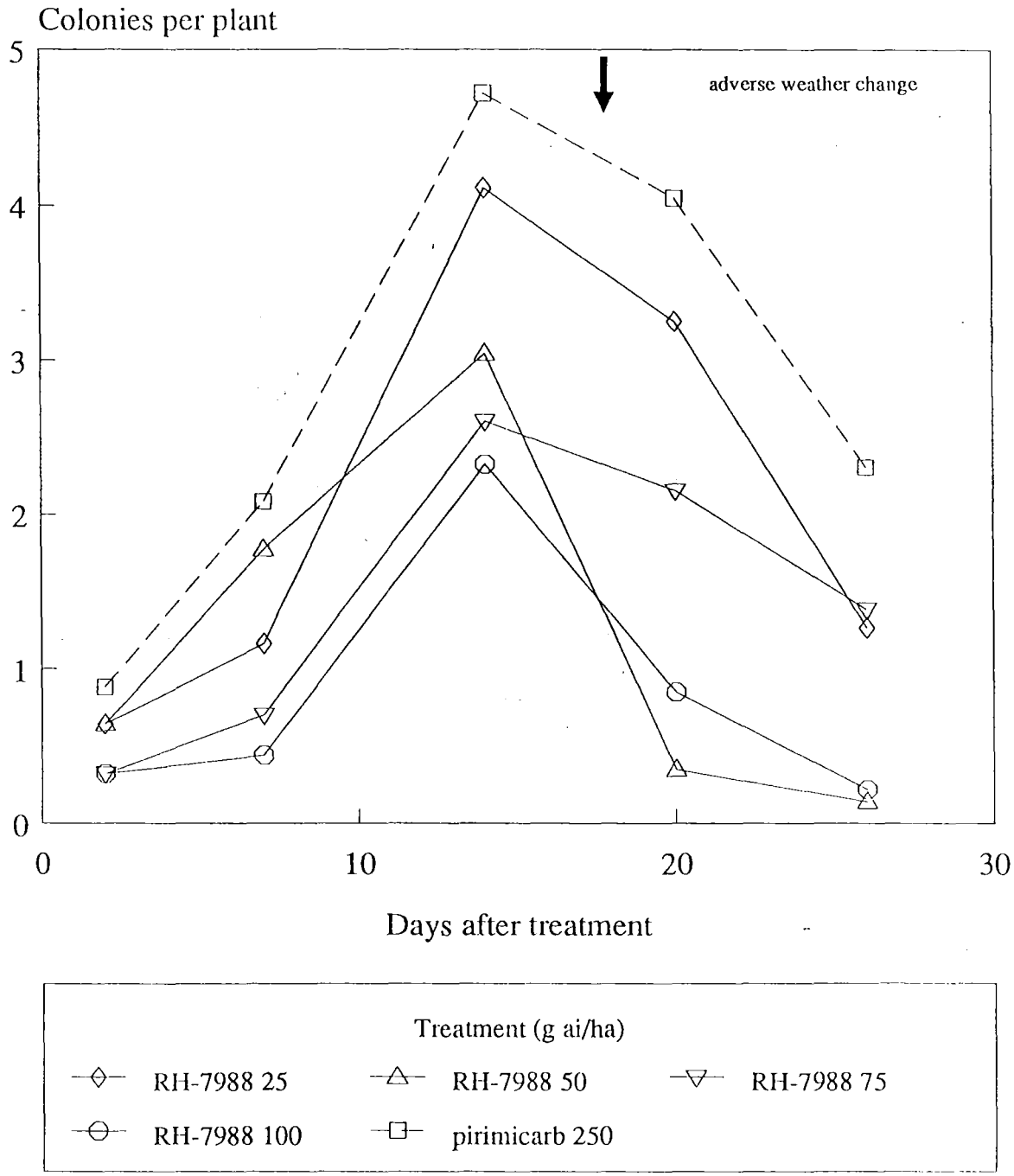

Figure 4.2b: The mean number of cabbage aphid colonies per plant on cabbages in Field Experiment III.

There were sufficient data to analyse the size of aphid colonies (no differentiation between species) for all sample dates except 2 DAT, but only after the number of size categories had been collapsed down to two (colonies composed of only one aphid and colonies composed of greater than one aphid). The results of the Chi-square analysis are presented in Table 4.6.

Treatment effects were significant 14 DAT only ($p < 0.05$). The trend shown in the results was similar to that indicated by the number of colonies per plant (Figs 4.2a and b), the size of the colonies increasing to 14 DAT and decreasing over the two following sample dates (20 and 26 DAT).

The number of alate aphids per plant (no differentiation between species) had a significant ($p < 0.05$) treatment effect only 2 and 7 DAT (Table 4.7). All RH-7988 treatments had significantly fewer alatae per plant than the pirimicarb treatment at both these sample dates, except RH-7988 at 50g a.i./ha 2 DAT.

Table 4.6: Chi-square analysis on the size of aphid colonies (Field Experiment III). Data grouped into two categories: single aphid colonies and those with more than one individual

Treatment		Days after treatment							
(g a.i./ha)		7		14		20		26	
		Size of colony							
		1	>1	1	>1	1	>1	1	>1
RH-7988	25	145	192	258	209	64	64	39	23
	50	105	171	174	219	50	50	39	29
	75	111	108	131	168	52	84	31	29
	100	59	66	124	133	25	44	30	18
pirimicarb	250	132	183	232	228	37	38	28	17
Chi-square		8.996		14.444		7.531		2.237	
p-value		0.061		0.006		0.110		0.692	
n		1272		1876		508		283	

Table 4.7: The number of alatae per plant (Field Experiment III)

Treatment		Days after treatment				
(g a.i./ha)		2	7	14	20	26
RH-7988	25	7.92	12.64	8.56	3.79	0.90
	50	9.28	10.47	9.12	3.20	0.90
	75	6.76	8.09	9.48	3.86	2.30
	100	6.36	7.88	9.36	0.93	1.30
pirimicarb	250	10.36	16.24	10.02	3.67	1.50
LSD		2.111	3.285	NS	NS	NS
SEM		0.754	1.173	0.823	0.928	0.457
n		125	125	125	50	50

Comparing the effect of RH-7988 at 100 g a.i./ha with and without the surfactant (Table 4.8) shows that the actual mean number of aphid colonies per plant were not very different. The range of data was greater in the treatment without surfactant, however, this may be due to the smaller sample size (3 plants/2 replicates cf. 5 plants/5 replicates). The untreated plots had markedly higher numbers of aphid colonies per plant but the difference was smaller in later samples.

Treatment had no significant effect on the yield of cabbages in this field experiment as measured by the three weights recorded (total fresh weight, market fresh weight and market dry weight). As shown in Fig 4.3, all error bars overlapped not only for the RH-7988 treatments but also for the pirimicarb, untreated and no-surfactant treatments.

Table 4.8: The number of aphid colonies per plant (observation plots, Field Experiment III)

Treatment (g a.i./ha)		Days after treatment					
		7		14		26	
cell content:	mean SEM	Aphid species					
		GPA	CA	GPA	CA	GPA	CA
RH-7988 100	+ surf	4.6	0.4	8.0	2.3	3.6	0.2
		1.13	0.32	1.00	0.54	0.93	0.43
	- surf	1.8	0.5	8.50	0.83	5.17	0.17
		1.17	0.84	7.48	0.75	1.47	0.41
untreated		27.2	5.7	28.8	5.0	8.5	4.2
		7.99	5.20	20.36	3.16	5.21	5.95

+ surf - Triton B1956 at 0.03% (a treatment in the main experiment)

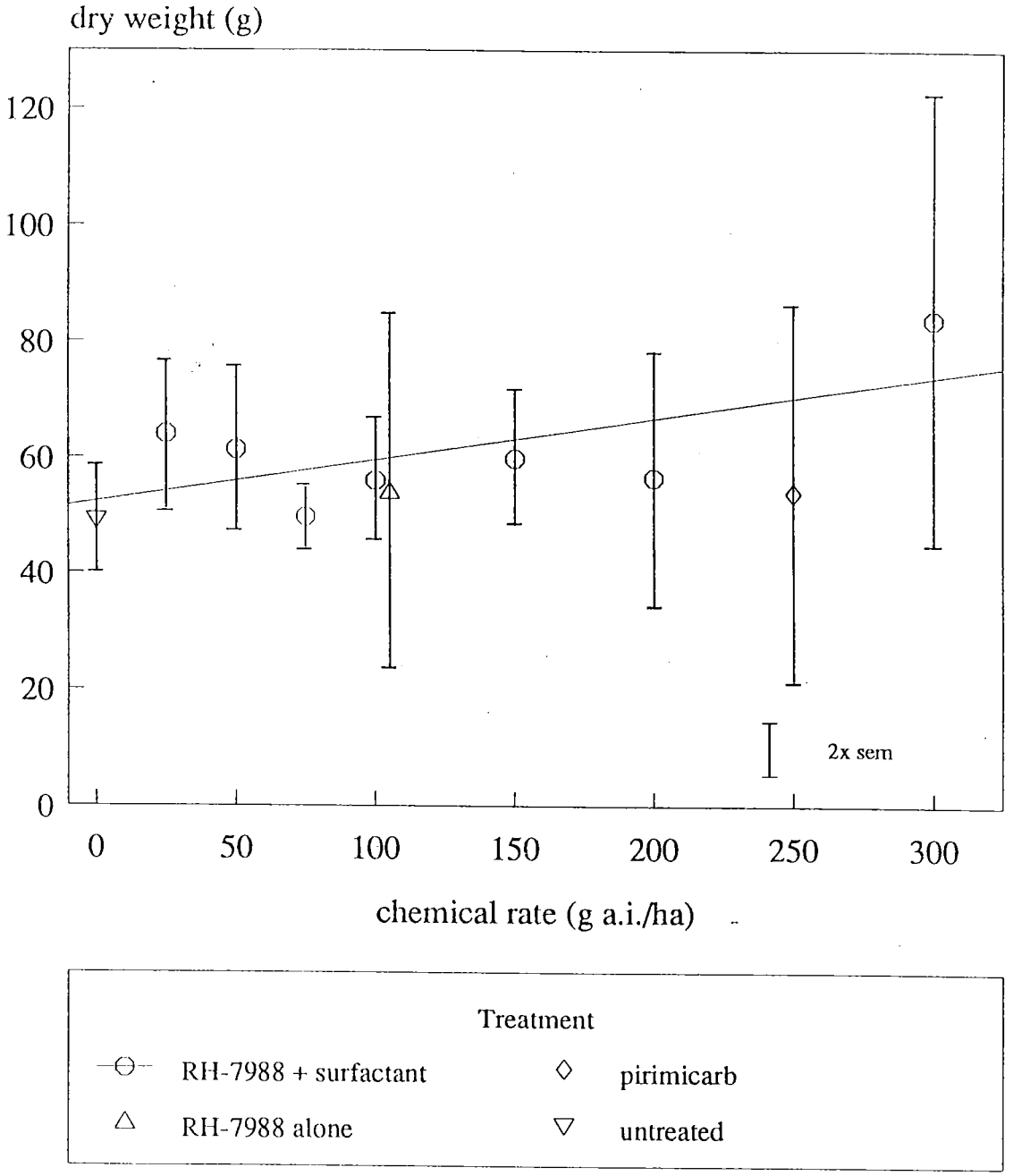

Figure 4.3: Dry weight yield of cabbages in Field Experiment III.

4.2 Laboratory experiments

4.2.1 LC_{50} determination

The LC_{50} values of RH-7988 and pirimicarb for green peach aphid were 2-fold and 4-fold greater than those for cabbage aphid (Table 4.9). Wide confidence intervals (95%) were recorded for all LC_{50} values, which were not significantly different to each other within a species. The slope of the regression lines of both aphicides were significantly different for green peach aphid but not for cabbage aphid.

Table 4.9: The LC_{50} values and gradients of the concentration-mortality lines for RH-7988 and pirimicarb on green peach aphid and cabbage aphid.

Species	Aphicide	LC_{50} (μg/l)	(95% fiducial limits)	Slope	SEM Slope
GPA	RH-7988	41.7	(25.66, 81.03)	1.4	0.22
	pirimicarb	73.5	(32.65, 169.75)	0.7	0.12
CA	RH-7988	19.0	(7.70, 64.45)	1.1	0.17
	pirimicarb	16.5	(5.88, 54.30)	0.9	0.11

4.2.2 Effect of post-treatment temperature on toxicity

The toxicity of RH-7988 and pirimicarb to both aphid species did not change significantly over the temperature range tested (Fig 4.4a and b). The 95% confidence intervals for the percentage of aphids that were moribund after 24 hours overlapped and the slopes of the fitted regressions were low. Regressions fitted were significant for cabbage aphid ($p<0.05$), but were not significant for green peach aphid ($p>0.05$).

The count of dead aphids 24 hours after aphicide application and the 48 hour counts (dead and moribund) did not have consistently smaller 95% CIs. The slopes of the regressions for moribund aphids after 48 hours were lower than those for the 24 hour data.

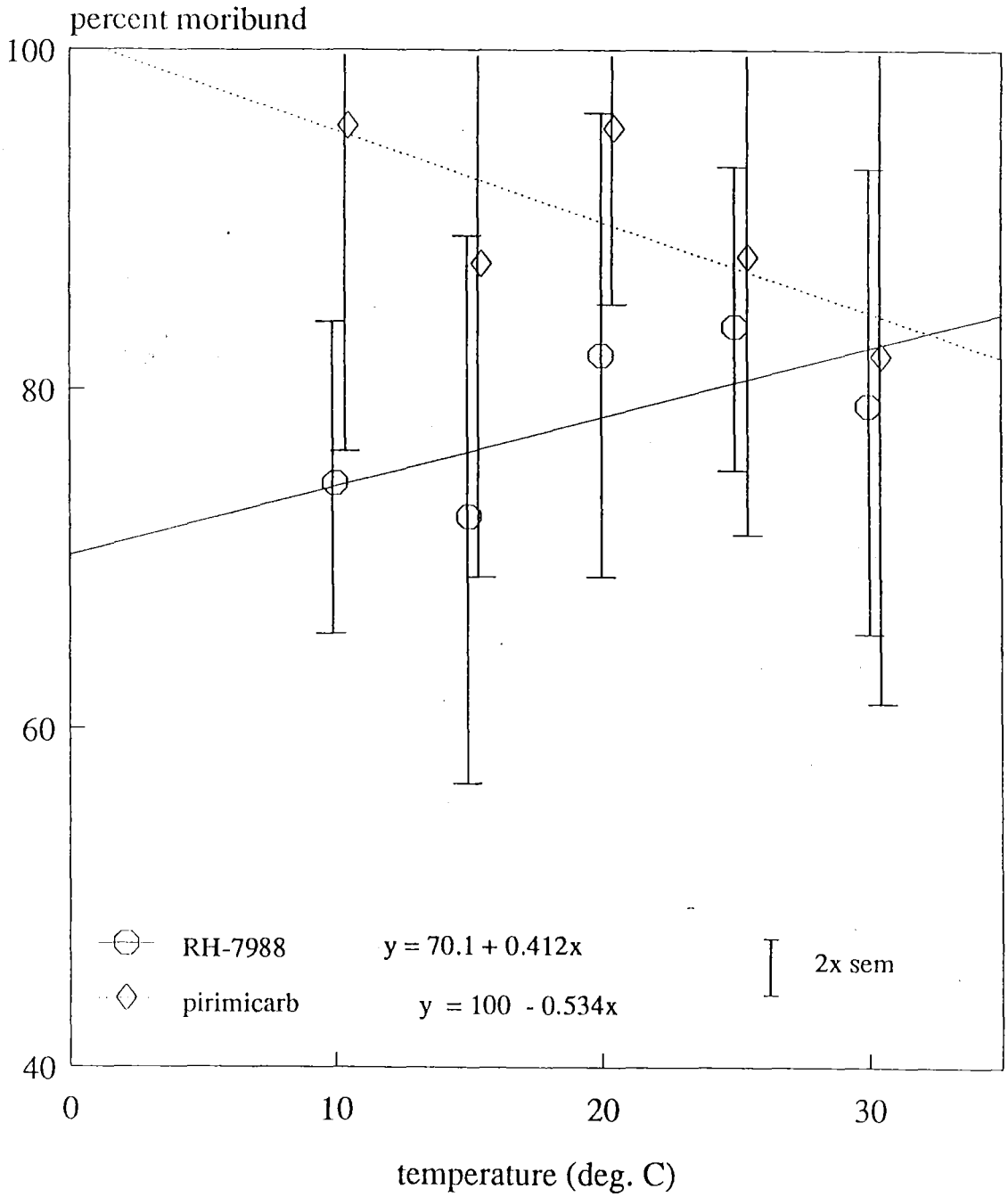

Figure 4.4a: The effect of temperature on the toxicity of RH-7988 and pirimicarb to green peach aphid.

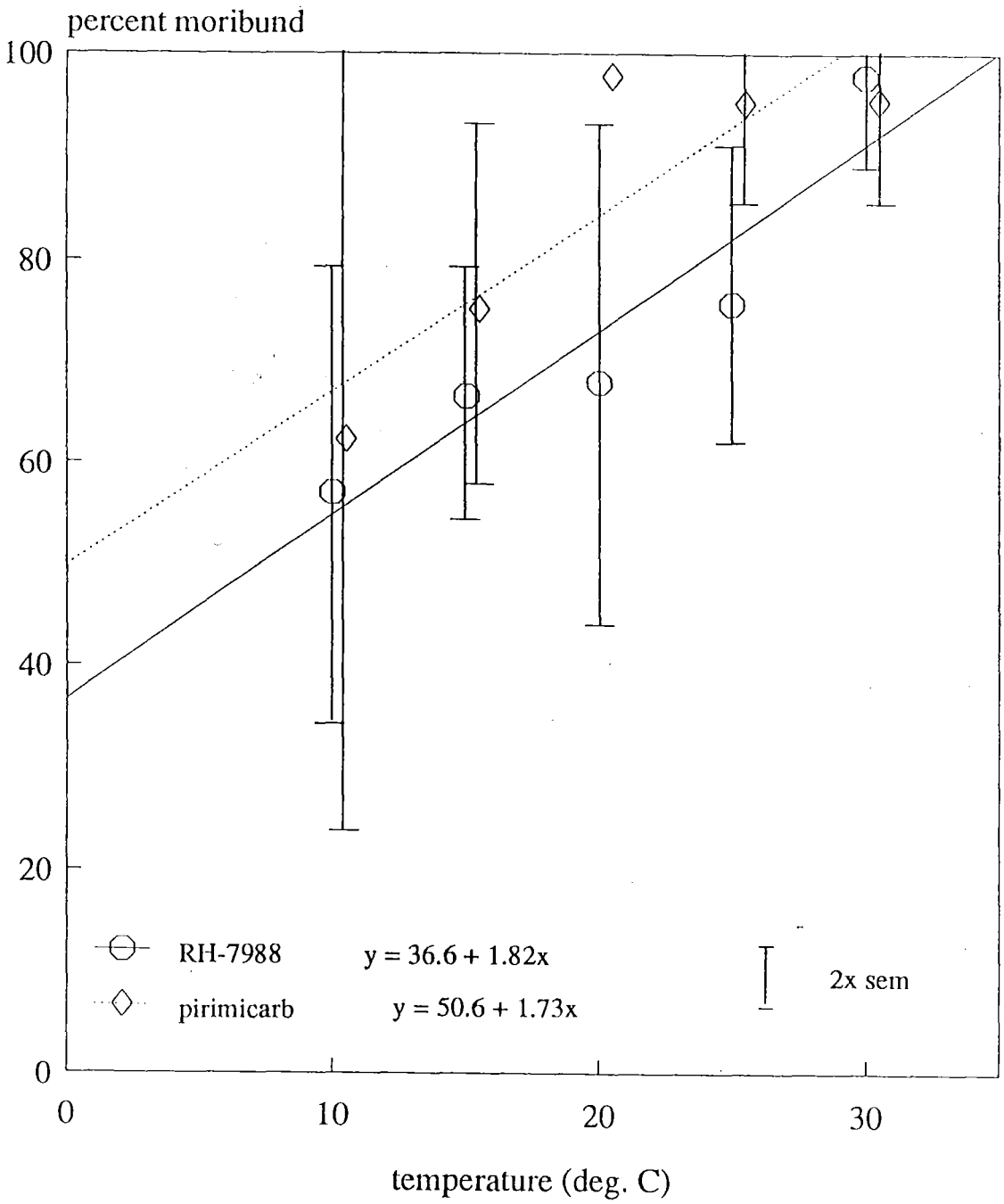

Figure 4.4b: The effect of temperature on the toxicity of RH-7988 and pirimicarb to cabbage aphid.

4.2.3 Residual activity against aphids

The aphicide treatments had a significant effect ($p < 0.05$) on green peach aphid up to 10 DAT and on cabbage aphid for only 5 DAT (Table 4.10a and b). Within these dates the only significant differences between RH-7988 and pirimicarb were at 1 DAT for cabbage aphid where RH-7988 at 50g and 75g a.i./ha had significantly fewer moribund apterae and at 10 DAT for green peach aphid at which time RH-7988 at 200g a.i./ha had significantly more moribund apterae than pirimicarb.

Additional counts were made on leaves that had emerged after the application of aphicides. These counts had significant treatment effects ($p < 0.05$) 23 and 27 DAT for green peach aphid and 18 DAT for cabbage aphid. RH-7988 at 100g and 200g a.i./ha had significantly more moribund aphids than pirimicarb 18 and 23 DAT (cabbage aphid and green peach aphid, respectively).

Table 4.10: Residual activity of RH-7988 (corrected means for % moribund) against green peach aphid and cabbage aphid in an insectary

(a) green peach aphid

Treatment (g a.i./ha)		Days after treatment 1	4	10	20	23*	27*
RH-7988	50	40.0	34.4	28.0	4.0	4.0	0.0
	75	72.0	81.0	21.4	0.0	10.0	26.0
	100	71.8	35.0	15.0	0.0	60.6	23.0
	200	80.0	73.0	90.0	0.0	55.0	30.0
pirimicarb	250	67.5	39.0	12.0	0.0	5.0	1.0
$LSD_{0.05}$		42.04	52.54	28.99	NS	27.37	30.72
SEM		12.60	15.74	8.68	1.51	8.20	92.0
n		34	34	34	34	34	34

(b) cabbage aphid

Treatment (g a.i./ha)		Days after treatment 1	5	10	20	18*
RH-7988	50	62.0	50.6	10.0	5.0	12.0
	75	63.0	15.0	20.0	0.0	26.0
	100	95.0	63.0	49.6	3.4	86.6
	200	91.0	78.0	23.4	6.6	64.2
pirimicarb	250	100.0	51.0	10.6	8.0	23.0
$LSD_{0.05}$		21.72	45.54	NS	NS	34.97
SEM		6.51	13.65	11.55	4.54	10.48
n		34	34	34	34	34

* - counts made on leaves that emerged subsequent to aphicide application

4.2.4 Toxicity to natural enemies

Neither species of ladybird were affected by RH-7988 at either rate tested (Table 4.11). The toxicity of RH-7988 was greater towards adult than third instar lacewing, although the former had a greater number of moribund individuals in the control treatment. The hymenopteran parasitoids were the most affected, this group comprised mainly of the primary parasitoid, *Diaeretiella rapae* but also included a hyperparasitoid, *Alloxysta infuscata*.

Table 4.11: Toxicity of RH-7988 to natural enemies of aphids

Species and lifestage		Rate (g a.i./ha)	Percent moribund	SEM	n
Tasmanian lacewing	larvae	0	12.4	2.33	25
		100	38.6	5.79	31
		200	43.3	7.31	30
	adult	0	42.5	8.25	9
		100	90.0	4.47	10
		200	90.0	4.47	10
eleven-spotted and orange-spotted ladybird	adult	0	0	0	8
		100	0	0	9
		200	0	0	10
Hymenoptera	adult	0	36.7	9.65	24
		100	96.0	1.39	25
		200	100.0	0	27

CHAPTER FIVE

DISCUSSION AND CONCLUSIONS

5.1 Field experiments

5.1.1 Results

The results of the field trials showed that initially RH-7988 is as effective as pirimicarb in controlling both green peach aphid and cabbage aphid in cabbage crops and that the action of RH-7988 against aphids appeared to persist longer than did the action of pirimicarb. The number of aphid colonies per plant did not attain levels equal to or greater than that of the control treatment within 35 days of RH-7988 application. The number of colonies of aphids per plant treated with the recommended field rate of RH-7988 did not exceed that of pirimicarb until 26 days after application.

These field experiments agree with what has been found overseas in other work. Murray *et al* (1988) reported that field experiments with RH-7988 applied to a range of aphid species and host crops showed that RH-7988 (concentrations varying from 35 to 140g a.i./ha) to have good knockdown of aphids two days after treatment and good residual control 7 to 18 days after treatment. Species and hosts included in these experiments were green peach aphid on sugar beet and cabbage aphid on Brussels sprout.

The use of RH-7988 did not appear to deter alatae from landing on the cabbages. Alate aphids were present on the cabbages throughout the experiments with significant differences between treatments only occurring two and seven days after treatment. Trumble (1982) consistently found alate aphids on broccoli in autumn and winter field experiments.

The effect of RH-7988 on the quantity of yield was inconsistent. In Field Experiment II the higher rates of RH-7988 significantly lowered the yield of cabbage heads, while in Field Experiment III there was no significant effect of RH-7988 on the cabbage yield. There was a wider range of RH-7988 concentrations in Field Experiment III as well as a wide variation as measured by the standard error of the mean. RH-7988 had no phytotoxic effect on the cabbages.

Management of the cabbages in the field experiments was not up to commercial standards. This was because the author had never grown cabbages before. Over the course of the three field experiments management did improve, however, the proportion of the yields in the "premium grade" were still lower than what would be expected commercially.

The availability of irrigation was limited as it was rotated in with the other crops on the research farm, for this reason water was not always applied as needed.

A large population of lepidopteran pests established in the third experiment (predominantly *Plutella xylostella* (L.)). Thuricide HP WP, (0.5-1.0 kg/ha, *Bacillus thuringiensis*) was applied on a regular basis (in all field trials) but failed to adequately control these pests (in this case). A synthetic chemical insecticide could have been used to gain better control but this may have had an effect on the aphid populations and so was not used. Observations indicated that the presence of these larvae was uniform over all treatments, although no samples were taken to estimate their populations.

Climatic variables may have exerted an effect on the field experiments on two occasions. In the second field experiment meteorological data indicated that on the day following treatment the minimum temperature increased, the relative humidity dropped drastically (to 28% from an average of 79% for the previous seven days), the wind was stronger (7.7m/s up from a 3.5m/s average for the previous seven days) and 9.3mm of precipitation was recorded. This may have contributed to the observed drop in the number of aphid colonies on the first sample after the treatments were applied.

Meteorological data for the third field experiment revealed that there was a cold and wet change in the weather 18 DAT (marked by an arrow on Figs 4.2a and b). Over a two day period, rainfall of 17mm was recorded and the mean daily temperature dropped from 15.6^{O}C over the previous 7 days to 10^{O}C. This inclement weather may have led to a decline in the aphid population of each treatment.

5.1.2 Sampling methods

To evaluate an aphicide for use in cabbage crops a sampling method similar to that used for monitoring aphid populations in commercial cabbage fields was considered to be the most useful. Theunissen (1989) developed a monitoring programme for cabbage aphid in three varieties of brassica in the Netherlands using the percentage of infested plants in a sample as the measuring unit. These were simplistic estimates in that a plant was infested regardless of the number of aphids on it and without an appreciation for the control exerted by the aphid's natural enemies.

A count of the number of aphid colonies per plant was relatively easy to achieve, although the loose aggregation of green peach aphid colonies (c.f. cabbage aphid colonies) made delineation of the colonies difficult at times and may have introduced some error.

Size of the aphid colonies was harder to determine. The time required to count the number of aphids in each colony would have been prohibitive, so an alternative was developed. The colonies were ranked into size categories depending on their estimated size. However, the data gathered were not adequate for Chi-square analysis in that there were too many zero counts in the data. To reduce the number of zeroes the number of categories was collapsed down. This went to the extent of having two size categories in Field

Experiment III, colonies with a single aphid and colonies with more than one aphid.

To avoid the problem with zero counts the data may be transformed. Hayman and Lowe (1961) suggest three transformations that could be made to cabbage aphid counts. In the analysis of whole plant counts the error variance increased with the mean in a log relationship and Hayman and Lowe suggested the transformations were required to stabilise the variance.

The numerical categories used were possibly not suitable for the work and a scale of the following categories: small, medium and large colonies may have worked better, especially if they were calibrated to the colony characteristics of each of the aphid species first. More operator error may be introduced in this method through looser definition of the categories.

An another way to get over the problem of zero counts may have been to remove them by omitting those leaves with no colonies on them from the analysis. Results would be presented as the number of colonies per plant and size of those colonies (rather than the size of colonies per plant).

Many authors have counted the total number of aphids per plant in their experiments, e.g., Heathcote *et al* (1969), Way *et al* (1969), and Dunn and Kempton (1971). Counting was generally done in the field but, as Way *et al*

(1971) noted, field counts underestimate aphid numbers on leaves (of Brussels sprout) and missed aphids and natural enemies in the sprouts.

To use a total count of aphids per plant in this project a number of changes would have been required. It would have taken longer to sample each plant and this may have required fewer samples per plot or reduced replication in the absence of more labour input. If plants were to be removed to count aphids in the laboratory, which would have been quite likely once the plants started forming their heads, larger plots would be needed to prevent any possible effects due to the loss of plants from the plots. These changes were not made and so the sampling method used was the best in the given situation.

In acknowledging the problems associated with counting aphid numbers in the field, raised by Way *et al* (1969), it was assumed that these were consistent over all treatments and therefore had no effect on the results.

5.2 Laboratory experiments

5.2.1 Aphids

The LC_{50} values determined for the two aphicides were not significantly different for either aphid species. There was wide variation in the responses of the two aphid species to the aphicides. This could possibly have been due to a number of factors:

(a) Testing was not restricted to one clone of each species. At the time of research controlled environment rooms were not available and the colonies were maintained in insectaries and were subjected to the outside environment. Growth of the colonies was not, therefore, optimal and populations from the field (from unsprayed areas) were required to provide the numbers for testing.

(b) Small groups of aphids were caged together in these tests and the aphids, especially green peach aphid, showed an ability to escape from the cages in which they were held. Very few aphids escaped from the control treatments. This suggests that the aphicides may have a irritant effect. Although 20 aphids were treated per aphicide application, these were split into four groups of five aphids caged on a leaf disc. The loss of one aphid in this situation had a greater proportionate effect on the results than larger groups (on larger leaf discs) would have had. The cage design could have been refined to reduce the numbers of aphids escaping.

(c) Experimental variables may have been inadequately controlled. If these variables were identified then the variation in results could be reduced, e.g., the humidity was not measured or controlled in any way and may have varied between runs of the experiment.

(d) The variation may have been non-genetic. The nutritional and/or the physiological status of the aphids may vary, especially when gathered from "wild" populations. Furk and Roberts (1985) reported a 2.8 fold variation in the LC_{50} in successive tests on cabbage aphids derived from a single female. They suggested that since the amount of wax produced by cabbage aphid varies considerably between individuals of a clone, this wax may affect insecticide penetration, and therefore, variation in the wax produced may contribute to the variation in response.

The slopes of the response curves for both chemicals and both species were relatively low (0.7-1.4). Baker (1978) tested a range of chemicals on a resistant glasshouse strain of green peach aphid and compared the results against a susceptible field strain. For pirimicarb the slope of the response curve of the resistant strain was much lower than that for the susceptible strain. It is possible that the aphids tested in these experiments had some degree of resistance (or tolerance) to the two aphicides, giving lower response curves. However, as Baker (1978) used a leaf-dip method, his results cannot be directly compared with this experiment.

The analysis of the 24 hour data rather than the 48 hour data was suggested by Rohm and Haas (NZ) Ltd. It was reasoned that this would give an indication of the rapidity of the action of RH-7988, the knockdown effect. As it was the 48 hour data was not adequate for analysis because the G value was to large.

This probably resulted because the aim was to determine the 24 hour LC_{50}. The FAO recommendations for pesticide testing on adult aphids (Anon, 1979) state that mortality counts should be done after 24 hours or more, according to the insecticide used.

By analysing the 24 hour (moribund) data, some indication of sub-lethal effects of the aphicides can be gained. For example, it was noted in the LC_{50} experiment that while some of those aphids in the control treatments were feeding and reproducing on the leaf discs after 24 hours, this observation was rare in aphids that were apparently unaffected by an aphicide treatment.

The use of 48 hour data gives the most precise results (Galley, 1968) and, in general, is the better data set to use. However, the variable analysed should be the one which best fulfils the requirements of the study.

One factor that may have had an influence in all the laboratory experiments was related to leaf discs. The plants from which leaf discs were taken were grown in a glasshouse and the leaf discs were punched from young fresh tissue. By comparison, the leaves from which the aphids came were often older and tougher leaves with thicker wax layers. The sudden change of substrate may have disturbed the aphids and upset their feeding therefore, introducing variability into the results.

The lack of effect of post-treatment temperature on the toxicity of RH-7988 to aphids are in agreement with the findings of McLeod (1987) for green peach aphid. However, McLeod found that pirimicarb had a positive temperature coefficient and the results in this trial did not indicate any significant relationship. However, the differences in experimental methods possibly explain the differences in results. In McLeod's experiments the green peach aphids used were not removed from the host substrate on which they were reared, and the aphicide solutions were equilibrated to the post-treatment temperature before the aphids were dipped into them.

A further factor influencing the outcome of the temperature-toxicity experiments may have been from the stress caused by a sudden change of temperature after treatment. At least, this may have compounded the stress imposed by the aphicide application on the aphids. Pre-conditioning of the aphids to the set post-treatment temperature may decrease the variability in results.

In a manner similar to that demonstrated in the field experiments, the residual activity trials illustrated that there was very little difference between RH-7988 and pirimicarb in controlling the two aphid species over the first 7 days after application. RH-7988 gave longer control over aphids than pirimicarb in the field experiments, but significant differences were not observed later in the residual activity trial, due to high inter-replicate variation (on leaves directly intercepting the aphicide).

Baranyovits (1969) reported that pirimicarb has a relatively short residual life as a spray deposit on plants (a half-life of less than 24 hours). Murray *et al* (1988) reported excellent residual activity of RH-7988 and poor residual activity of pirimicarb against green peach aphid on broccoli (91% and 21% mortality, respectively).

Counts on leaves that emerged after treatment indicated that RH-7988 had superior systemic activity than pirimicarb and gave good control in the apical meristem of the cabbages three weeks after application. However, these results do not comprehensively prove systemic activity of RH-7988 in cabbages. Galley (1968) and Matthews (1984) describe experiments that could be set up to further study this area.

In experiments by others, RH-7988 has exhibited both upward and downward translocation in the plant and can control root-feeding aphids by a foliar application as well as leaf-feeding aphids by soil application (Anon, 1989). Murray *et al* (1988) reported excellent systemic properties of RH-7988 on tobacco against green peach aphid when applied as a soil drench. Pirimicarb, on the other hand, exhibits systemic activity up the plant only (Baranyovits, 1969).

5.2.2 Natural enemies

The data presented for this experiment is weak in strength. Numbers tested were too low as was the number of replications. However, from the data gathered (and presented) ladybird adults were the least affected natural enemy, followed by lacewing larvae, adults and hymenopteran parasitoids.

The concentration of RH-7988 applied to the natural enemies in this experiment killed 100 percent of both aphid species in earlier work. The corrected mortalities for the predators are all less than 50 percent, but the parasitoids appear to be susceptible.

Murray *et al* (1988) tested RH-7988 (3.82 $\mu g/cm^2$) on six beneficial insect species. Mortality was less than 30% for all of the species, except one hymenopteran parasitoid which had a mortality between 31 and 70%.

There was a high proportion of moribund individuals in the control groups of both the lacewing adults and Hymenoptera (42.5% and 36.7%, respectively) which resulted in lower corrected percentages of moribund insects in the insecticide treatments. It is possible that this was caused by the insects being exposed to too much carbon dioxide for too long when they were anaesthetised.

Baranyovits (1969) noted that in both field and laboratory experiments pirimicarb was relatively harmless to most aphid predators and parasites (hoverflies being an exception). Proctor and Baranyovits (1969) reported that pirimicarb had very little effect on four predator species of green peach aphid. Their work also showed that pirimicarb did not impede adult parasitoid emergence from treated pupae and fresh deposits of pirimicarb caused only 15% mortality of the parasitoid adults.

Helgesen and Tauber (1974) found that pirimicarb was not toxic to the life stages of three beneficial insects. Syrett and Penman (1980) reported a 1000-10 000 fold difference in the toxicity of pirimicarb to aphids as compared to two predator species (Tasmanian lacewing and 11-spotted ladybird).

Considering the literature and this experiment, RH-7988 appears to be as selective towards aphids as is pirimicarb. This experiment would have benefited from greater replication and uniformity of materials, but the best comparison would be achieved by testing the two aphicides in the same trial.

5.3 Conclusions

At the recommended field rate, RH-7988 gave excellent control of green peach aphid and cabbage aphid, both in the laboratory and in the field. The response of natural enemies to RH-7988 varied, but in general the aphicide was selective towards the aphids.

Good control of aphids was achieved with RH-7988 on potted plants for 20 days after treatment, both on leaves sprayed with RH-7988 and leaves not sprayed with RH-7988.

In the field, RH-7988 was as effective as pirimicarb in controlling aphids over the short term (up to 14 days after treatment) and superior to pirimicarb over a longer time period (up to 35 days after treatment). RH-7988 was not phytotoxic to the cabbages, alone or in combination with surfactants.

RH-7988 is an aphicide that can be used in the present spraying regime in New Zealand brassica crops and, more importantly, can potentially be used in IPM programmes that may be subsequently introduced for both forage brassicas and vegetable brassicas.

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